

Evaluation of the suitability of electronic nose based on fast GC for distinguishing between the plum spirits of different geographical origins

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Received: 2 September 2015 / Revised: 8 March 2016 / Accepted: 30 March 2016 / Published online: 16 April 2016
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Abstract Plum spirits are obtained exclusively via the alcoholic fermentation and distillation of meaty plums or must composed of plums, with or without stones. The European countries such as Poland, the Slovak Republic, the Czech Republic, Bulgaria and Romania are the leading producers of this alcoholic beverage. Information about the authentication of plum spirits produced in different countries can be found in the published literature. Gas chromatography, sensory analysis and spectroscopic techniques were the most frequently used research tools in those studies. So far, there were no available published reports about the suitability of an electronic nose based on fast GC for analyzing plum spirits. The volatile fractions of six samples of four different origins (Poland, Czech Republic, the Slovakia and Romania) with 40 % alcoholic content were analyzed using electronic nose based on fast GC (Heracles II). Four data analysis methods were used, namely principal component analysis, discriminant function analysis, soft independent modeling of class analogies and statistical quality control. In this work, for discrimination of results was used Euclidean distance between each sample, standard deviation and coefficient of variation. Conducted research was aimed at the evaluation of the suitability of an electronic nose based on fast GC for distinguishment between the volatile profiles of plum spirits of different geographical origins. These studies are the foundation for further work on discrimination of samples of plum spirits produced within a greater number of geographical regions.

All the applied chemometric methods turned out to be effective.

Keywords Electronic nose · Fast GC · Plum spirits · Authenticity test · Geographical origin

Introduction

In Europe, 46 traditional alcoholic beverages have been classified and defined, which includes inter alia, brandy, gin, liqueurs and fruit spirits [1]. Fruits spirits have been defined in the EU regulation as spirit drinks obtained exclusively via the alcoholic fermentation and distillation of meaty fruits or must composed of fruits, berries or vegetables, with or without stones. The amount of volatile substances, which has also been defined, must not exceed 200 g/hl of 100 % (v/v) alcohol. The content of alcohol derived from fruits must not be less than 37.5 % (v/v). Fruit spirits cannot be artificially flavored [2]. The name of a given fruit spirit should contain the name of the fruit from which it has been made, e.g., cherry spirit, or “Kirsch,” pear spirit, and plum spirit, or slivovitz [3].

Plum spirit is considered the most popular fruit spirit. The European countries such as Poland, the Slovak Republic, the Czech Republic, Bulgaria and Romania are the leading producers of this alcoholic beverage. Depending on the country of origin, plum spirit is known under different names, i.e., *śliwowica* (Poland), *slivovice* (the Czech Republic), *tuica* (Romania) and *slivovica* (the Slovak Republic) [3, 4]. Regardless of the country of origin of plum spirit, its production consists of the following steps: washing and pitting of plums (due to the legal regulations on the content of ethyl carbamate, a carcinogen present in the plum pits, the pits have to be removed), mashing and

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fermenting the plum purée. The next stage is distillation which is performed in order to obtain pure spirit. The heads and tails of the distillation run are not used in the production of this alcoholic beverage. Next, the main fraction is subjected to aging. Depending on the recipe, the obtained beverage is stored in glass containers or oak barrels for a period of time ranging from 2 to 5 years [5, 6]. Additionally, in the final production stage, the plum spirit can be diluted with water to achieve the alcohol content in the range of 40–50 % [3, 4].

Information about the authentication of plum spirits produced in different countries can be found in the published literature. A comparative sensory evaluation and GC–MS analysis were performed on the Polish plum spirit (i.e., Śliwowica Łącka), Slovakian Pravá Bošacka Slivovica and Romanian Tzuica z Clej-Napoca [4]. Fruit spirits produced from Serbian plums were also analyzed [7–9]. The Serbian plum spirits were studied in order to, inter alia, determine the content of volatile compounds in different kinds. The composition of four plum spirits, i.e., Manastrila from Belgrade, homemade Sokolova rakija, Karanka from the Užice region and Valjevka from the Valjevko region, was analyzed and compared by means of GC–FID and sensory analysis [7]. The Serbian plum spirits were also analyzed for the presence of Cu, Fe, Zn, Mn, Ca, Se and As using atomic absorption technique [9]. It was demonstrated that the composition of plum spirits is influenced by their geographical origin, preliminary processing, and the type and time of storage of the fruits [3, 4, 7, 9, 10]. Based on the cited publications, it is apparent that there are differences between the plum spirits originating from different countries in relation to the composition of volatile fraction and organoleptic properties. Gas chromatography (GC–MS [4, 11], GC–FID [7]), sensory analysis [4, 7, 11] and spectroscopic techniques were the most frequently used research tools in those studies. However, a comparison of volatile profiles of plum spirits from different countries obtained by means of an electronic nose is still lacking.

The electronic nose has been used for many years to authenticate alcoholic beverages. To this end, wines [12–21], tequila, whiskey, vodkas [22, 23] and Chinese spirit [24] were analyzed. Until now, the application of electronic nose was based on sensors, e.g., conductometric, amperometric and piezoelectric sensors [25]. However, the publications dealing with the authentication of alcoholic beverages by means of electronic nose based on fast GC are becoming more frequent lately. This specific type of electronic nose generates peaks with the highest discriminatory power, which produce a characteristic volatile profile, a so-called fingerprint, when exposed to the gas mixture [25]. The electronic nose based on fast GC was used to analyze inter alia whiskey [26, 27], wines [28] and liqueurs [29, 30].

Based on a review of published literature, it was concluded that an electronic nose based on fast GC has not yet been applied to investigate the volatile profiles of fruit spirits. Thus, it is necessary to broaden the knowledge about the use of this device for determining the volatile profiles of plum spirits originating from different countries. The main research aim was to evaluate whether the aforementioned electronic nose is suitable to distinguish between the plum spirits of different geographical origins. In addition, four statistical methods, namely principal component analysis (PCA), discriminant function analysis (DFA), soft independent modeling of class analogies (SIMCA) and statistical quality control (SQC), were used to analyze the data.

Materials and methods

Six plum spirits produced in four different countries located in Central and Eastern Europe, i.e., Poland (Śliwowica Łącka), the Czech Republic (Slivovice Zlatá R. Jelinek—CSZ, and Slivovice R. Jelinek—CS), the Slovak Republic (Zariecka Borovicka—SZB and Maraska Sljivovica—SM) and Romania (Țuica de Maramureș), were analyzed. All sampled plum spirits were purchased in 2013 and stored in tightly closed bottles at room temperature. The alcohol content of all beverages was 40 % v/v.

Prior to analysis, samples containing 2 ml of plum spirit and 4 ml of deionized water were prepared in 20 ml vials. Next, all samples were tightly closed with caps made of 20 mm thick PTFE/silicone membrane. High-purity deionized water used for diluting the plum spirits was obtained from a MilliQ A10 Gradient/Elix System (Millipore, USA).

In this study, the electronic nose based on fast GC (Heracles II, Alpha M.O.S., France) and equipped with two capillary columns of different polarity, i.e., non-polar MTX-5 (5 % diphenyl) and medium polarity MXT1701 (14 % cyanopropylphenyl) with dimensions of 10 m × 0.18 mm × 0.4 μm, and two ultrasensitive flame ionization detectors (μ-FIDs), was used. The electronic nose was also equipped with an autosampler (Odour Scanner HS 100; Gerstel, Germany). Each sample was incubated for 20 min at 40 °C, while the agitation speed was maintained at the level of 500 rpm. After incubation, only the headspace phase (2500 μl) was transferred into the injector heated to 200 °C. The chromatographic program consisted of the following parameters: initial temperature of 70 °C (kept for 18 s), then ramped at 2 °C/s to 270 °C (kept for 30 s) and the detector temperature of 270 °C. Hydrogen N5.0 was used as a carrier gas (Linde Gaz, Poland). Each sample was analyzed six times.

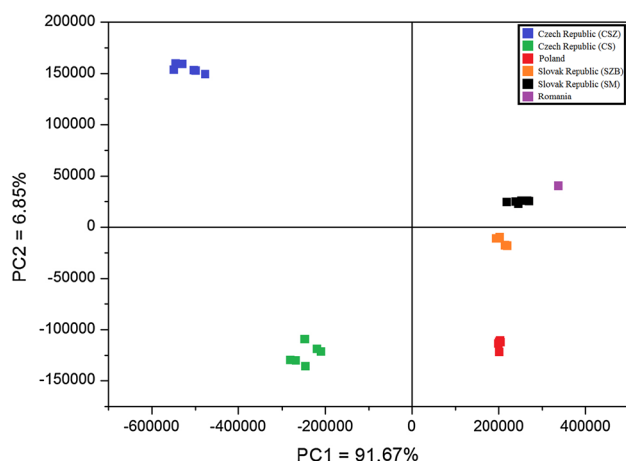


Fig. 1 The results of PCA 2D applied to plum spirits originating from different geographical regions; origin of samples was highlighted in: *purple color* Romania, *red color* Poland, *green color* Czech Republic (CS), *blue color* Czech Republic (CSZ), *orange color* Slovak Republic (SZB), *black color* Slovak Republic (SMB), and six repetitions of all types of samples were done

Four statistical methods were used to analyze the data, i.e., principal component analysis (PCA), discriminant function analysis (DFA), soft independent modeling of class analogies (SIMCA) and statistical quality control (SQC) by employing Alpha Soft software (V12.4, Alpha M.O.S, Toulouse, France). The *p* value was calculated by

using MANOVA algorithm. Figures were performed by using OriginPro 8.0 and Alpha Soft software.

Results and discussion

As there were no available published reports about the suitability of an electronic nose based on fast GC for analyzing plum spirits, four statistical methods were used to analyze the obtained data. The group consists of six repetitions of one sample.

PCA was the first employed chemometric method, which enabled a graphical representation of multidimensional relationships and the reduction in the number of variables [29]. In Fig. 1, the results of PCA 2D performed on the data collected by means of an electronic nose are presented, and the data points correspond to specific groups of plum spirits. Discrimination index of PCA 2D equals 91. The sum of the first and second principal components accounted for 98.52 % of data variance. PC1 accounted for 91.67 % and PC2 accounted for 6.85 %. It is apparent that the plot can be divided into two parts. The points corresponding to plum spirits from Poland, Slovak and Romanian are in the first and fourth quadrant of the plot. All calculated Euclidean distances and coefficient of variation by program Microsoft Excel 2007 are presented in Table 1. The distance between Romanian and Slovak Republic groups is higher than the distance between two Slovak

Table 1 Calculated values of Euclidean distances and coefficient of variation between groups according to PCA and DFA results

Relation between group		PCA 2D		PCA 3D		DFA		DFA 3D	
		E. distance	CV	E. distance	CV	E. distance	CV	E. distance	CV
Czech Republic (CSZ)	Czech Republic (CS)	389,901.67	0.05	389,908.92	0.05	854.03	>0.01	934.59	>0.01
Czech Republic (CSZ)	Romania	881,791.58	0.01	882,665.48	0.01	1746.49	>0.01	1746.63	>0.01
Czech Republic (CSZ)	Poland	721,143.65	0.02	774,346.13	0.02	2320.26	>0.01	2321.97	>0.01
Czech Republic (CSZ)	Slovak Republic (SZB)	742,250.85	0.02	742,375.69	0.02	1774.57	>0.01	1785.04	>0.01
Czech Republic (CSZ)	Slovak Republic (SMB)	776,962.63	0.03	777,471.84	0.03	1765.50	>0.01	1772.99	>0.01
Czech Republic (CS)	Poland	447,156.21	0.03	457,448.23	0.03	1581.30	>0.01	1607.79	>0.01
Czech Republic (CS)	Slovak Republic (SZB)	463,930.05	0.03	464,205.61	0.03	1152.28	>0.01	1167.29	>0.01
Czech Republic (CS)	Slovak Republic (SMB)	515,416.07	0.04	516,059.16	0.04	1168.22	>0.01	1188.16	>0.01
Czech Republic (CS)	Romania	359,988.56	0.01	395,873.92	0.01	1183.19	>0.01	1249.62	>0.01
Poland	Slovak Republic (SZB)	100,580.64	0.06	150,892.72	0.06	621.63	>0.01	630.29	>0.01
Poland	Slovak Republic (SMB)	145,981.61	0.08	162,215.76	0.08	663.86	>0.01	667.96	>0.01
Poland	Romania	218,332.22	0.01	258,361.29	0.01	728.51	>0.01	736.98	>0.01
Slovak Republic (SZB)	Slovak Republic (SMB)	57,300.15	0.24	70,895.49	0.24	60.05	0.02	67.22	0.01
Slovak Republic (SZB)	Romania	151,929.20	0.03	162,535.72	0.03	166.47	0.01	255.72	0.01
Slovak Republic (SMB)	Romania	109,448.85	0.02	128,538.24	0.02	77.97	0.02	201.00	0.01

E-distance—calculated Euclidean distance between two groups

CV (coefficient of variation)—sum of SD from two groups divided by Euclidean distance between them

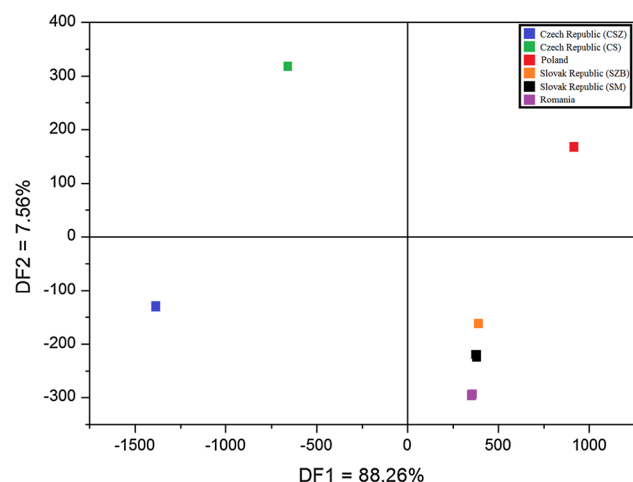


Fig. 2 The results of DFA 2D applied to plum spirits originating from different geographical regions; origin of samples was highlighted in: *purple color* Romania, *red color* Poland, *green color* Czech Republic (CS), *blue color* Czech Republic (CSZ), *orange color* Slovak Republic (SZB), *black color* Slovak Republic (SM), and six repetitions of all types of samples were done

Republic groups; additionally, the distances between Polish plum spirit group and other Slovak groups are also higher.

On the other hand, the points corresponding to two Czech Republic groups are located in second and third quadrant of the plot. These groups have largest distances between each other and other groups. It suggests that these groups differ the most from other groups, those originating from the Slovak Republic, Poland and Romania. The large distance is between points corresponding to Czech Slivovice Zlatá R. Jelinek (CSZ) and Slivovice R. Jelinek (CS), and different production process might be the reason of this observation. Czech plum spirit (CSZ) is aged in oak barrels, and it should be mentioned that the points corresponding to this group were located at the farthest distance from other spirit groups. Unfortunately, the use of PCA 2D does not allow for a clear distinction of plum spirits between their geographical origins based only on PC1 or PC2, but it allows for differentiation between groups.

DFA is a multivariate technique for describing a mathematical function, and it allows to distinguish among predefined groups of samples [25]. The DFA 2D is shown in Fig. 2. The DFA 2D method identifies DF1 and DF2 which explain 88.26 % and 7.56 % of the variability in the initial data set and which are represented as the axes of the DFA plot. As in the case of PCA 2D, the points corresponding to Czech plum spirits are separated by a large distance from the other samples. Moreover, the points belonging to the Czech (CSZ) group are located in the second and third quadrant of the plot and are clearly separated from the rest of the analyzed plum spirits. As with the results of the PCA, the distance between the Romanian and Slovakian

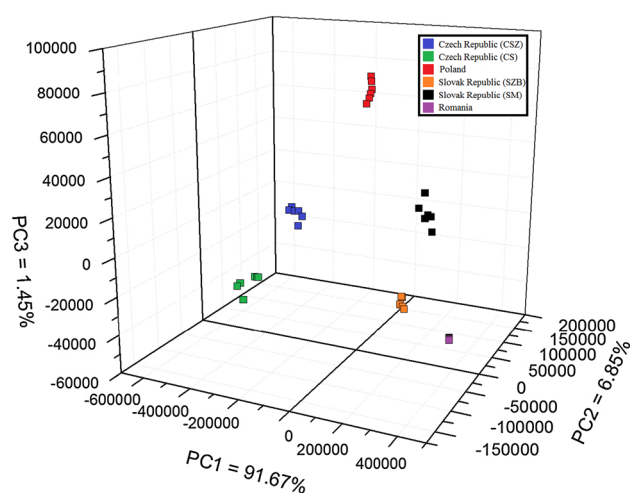


Fig. 3 The results of PCA 3D applied to plum spirits originating from different geographical regions; origin of samples was highlighted in: *purple color* Romania, *red color* Poland, *green color* Czech Republic (CS), *blue color* Czech Republic (CSZ), *orange color* Slovak Republic (SZB), *black color* Slovak Republic (SM), and six repetitions of all types of samples were done

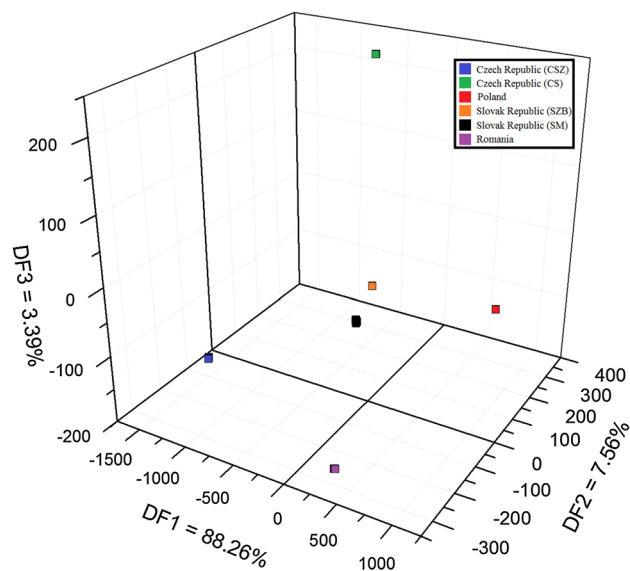
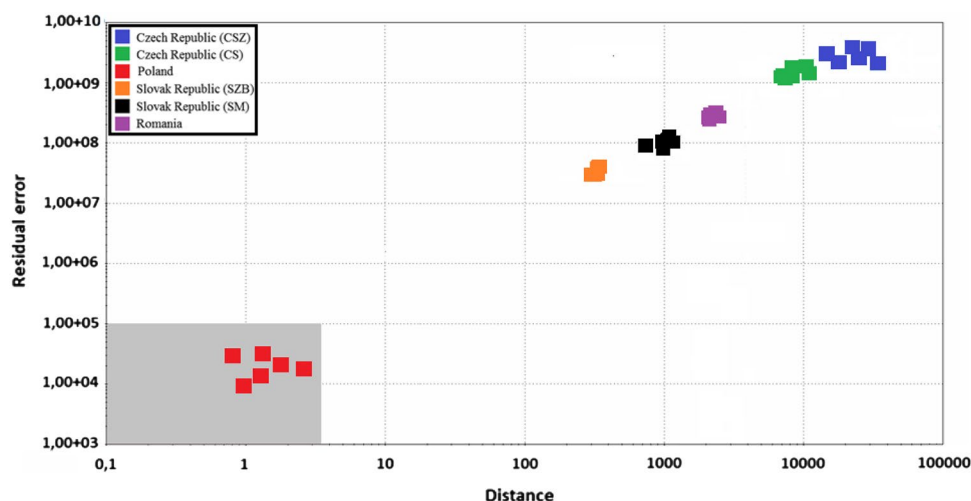


Fig. 4 The results of DFA 3D applied to plum spirits originating from different geographical regions; origin of samples was highlighted in: *purple color* Romania, *red color* Poland, *green color* Czech Republic (CS), *blue color* Czech Republic (CSZ), *orange color* Slovak Republic (SZB), *black color* Slovak Republic (SM), and six repetitions of all types of samples were done

groups is higher than the distance between two Slovakian groups. In comparison with the PCA 2D results, the points belonging to slivovitz are less dispersed. In the first quarter occur only points corresponding to a group of Polish plum spirit. However, the fourth quadrant includes only groups of Slovak beverages and plum spirit from Romania. For

Fig. 5 The results of SIMCA applied to plum spirits originating from different geographical regions; origin of samples was highlighted in: *purple color* Romania, *red color* Poland, *green color* Czech Republic (CS), *blue color* Czech Republic (CSZ), *orange color* Slovak Republic (SZB), *black color* Slovak Republic (SM), and six repetitions of all types of samples were done. The plum spirit originating from Poland is a reference group



this reason, based on DFA 2D plot, it is apparent that the DF1 enables distinguishing between the plum spirits of different geographical origins.

In order to verify the results obtained using two-dimensional PCA and DFA, three-dimensional analysis was performed (Figs. 3, 4). Figure 3 shows PCA 3D results, where PC1, PC2 and PC3, respectively, contributed to 91.67 %, 6.85 % and 1.45 % of the total variance. In Fig. 3, the Euclidean distance between two groups of plum spirits, Polish and Slovakian (SZB), is greater by about 50 % than in PCA 2D. In case of DFA 3D, the distance between Slovakian (SM) and Romanian is much greater, about 158 % of the distance in the DFA 2D plot. The distances between Slovak Republic (SZB) and Romanian was about 54 % greater in DFA 3D. Also in obtained results, it can be observed slightly smaller distance differences (over 15 %) between groups of drinks from Slovakia and Romania. This is caused by the third value (z-coordinate). The reduction to only two dimensions resulted in a loss of information, so the use of statistical methods in three dimensions is always beneficial to final results. Both graphs show, despite variations in the distance between the groups, that it is possible to clearly distinguish groups from each other, which leads to the distinction of geographical origin. In every case of distinction between groups, determination of the p value proved the null hypothesis to be true, which leads to the conclusion that the groups are uncorrelated.

In order to evaluate the suitability of an electronic nose for distinguishing of plum spirits of different geographical origins, the obtained data were analyzed by the SIMCA and SQC.

The SIMCA classification was used to compare all data to one group [31]. Analyses were performed using a classifier SIMCA in relation to the reference group:

Polish and Czech (CSZ). The area corresponding to the given class is shown on each plot as the gray rectangle in the left corner. In Fig. 5, validation score is 99, and thus, a significant distinction between Polish and other plum spirits groups was obtained. As in the case of plots in Figs. 1, 2, 3 and 4, the points corresponding to Polish plum spirit are in greater distance to other spirit groups. As in the case of other employed chemometric methods, the points corresponding to two plum spirits from Slovak Republic were closely located. In addition, it should be noted that the points corresponding to the Czech slivovitz are the most distant from the reference group. In Fig. 6, validation score is 97, and thus, a significant distinction between Czech (CSZ) and other plum spirits groups was obtained. Points corresponding to Czech (CS) were at the smallest distance from the gray area of confidence envelope. The distance between the reference group and the group corresponding to Czech (CS) was the smallest compared with the distances to other beverages.

In order to confirm the results obtained by SIMCA classifier, SQC was used. Figure 7 shows results of the SQC depending on Czech (CS) as reference group. The distance between Czech plum spirits is smaller than to other beverages. The largest distances are between Polish and Czech spirits and other spirits. Using the obtained results, it can be concluded that it is possible to distinguish Polish from Czech, Romanian and Slovakian plum spirits. In addition, it should be noted that the groups belonging to Slovakia are not far from each other. The Romanian group is also present in short distance away from them. For this reason, it can be concluded that the volatile profiles of Slovak and Romania are not significantly different, but they are still distinguishable.

Fig. 6 The results of SIMCA applied to plum spirits originating from different geographical regions; origin of samples was highlighted in: *purple color* Romania, *red color* Poland, *green color* Czech Republic (CS), *blue color* Czech Republic (CSZ), *orange color* Slovak Republic (SZB), *black color* Slovak Republic (SM), and six repetitions of all types of samples were done. The plum spirit originating from Czech Republic (CSZ) is a reference group

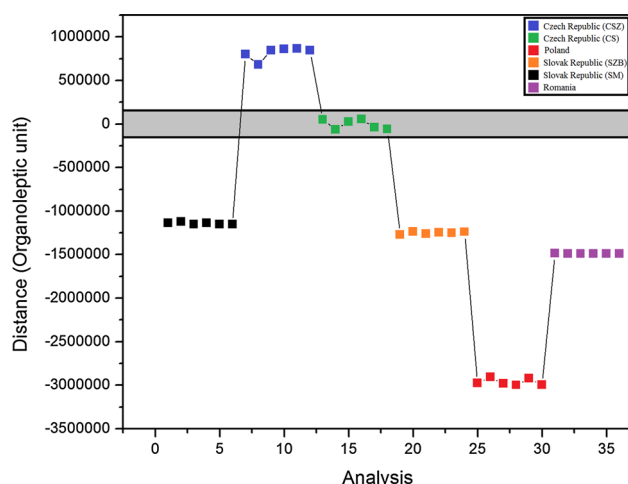
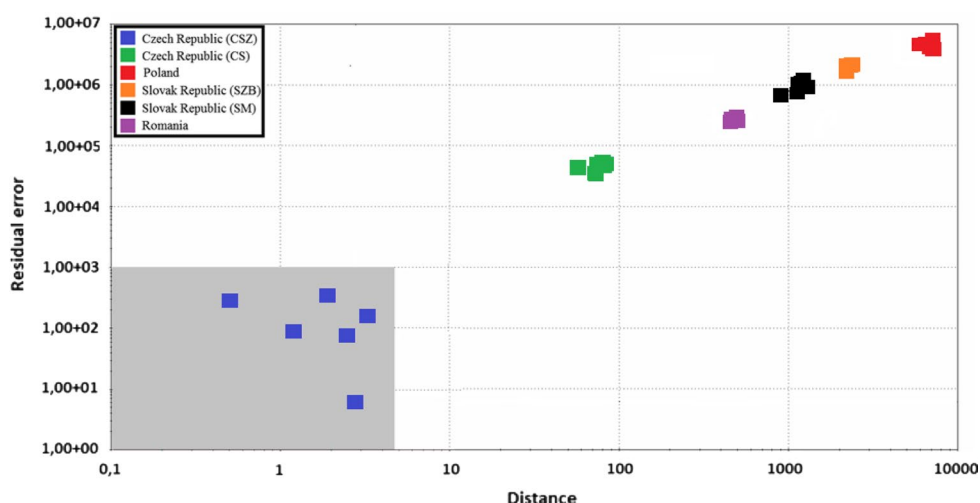


Fig. 7 The results of SIMCA applied to plum spirits originating from different geographical regions; origin of samples was highlighted in: *purple color* Romania, *red color* Poland, *green color* Czech Republic (CS), *blue color* Czech Republic (CSZ), *orange color* Slovak Republic (SZB), *black color* Slovak Republic (SM), and six repetitions of all types of samples were done. The plum spirit originating from Czech Republic (CS) is a reference group

Conclusion

This work reports, for the first time, the evaluation of an electronic nose based on fast GC for distinguishing between the volatile profiles of plum spirits of different geographical origins. It has been shown that it is possible to distinguish all analyzed groups of beverages. The electronic nose has shown that the highest distinction is between drinks from the Czech Republic and Poland. In addition, it has been shown that despite rather small differences between plum spirit from Slovak Republic and Romania it is still possible to distinguish them. All the applied chemometric methods turned out to be effective.

Analyses by PCA and DFA were done in two and three dimensions and demonstrated the ability to distinguish between all the groups of beverages. However, in the case of PCA and DFA, statistical methods in three dimensions should be used, because it has in all cases been beneficial to final results. Additionally, an analysis of the data using SIMCA and SQC was done in order to check the possibility of distinction of plum spirits according to their geographical origin. Moreover, despite the differences in the production of samples from Czech Republic, electronic nose has allowed for the distinction of samples from other beverages depending on the origin.

The positive results of the research have the potential to broaden the scope of scientific research in the use of electronic nose to investigate the geographical origin of plum spirits. These studies are the foundation to further work on discrimination of samples of plum spirits produced within a greater number of geographical regions. In the future, these tests may be used to study not only the geographical origin of fruit spirits, but also other spirit beverages.

Acknowledgments The authors acknowledge the financial support for this study by Grant No. 2012/05/B/ST4/01984 from the National Science Centre of Poland.

Compliance with ethical standards

Conflict of interest None.

Compliance with ethics requirements This article does not contain any studies with human or animal subjects.

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